

II. AMENDMENTS TO THE SUBSTITUTE SPECIFICATION.**1. At page 1 of the CIP Replacement Application****Title Title**

An Apparatus and Method And Techniques for Measuring and Correlating
Characteristics of Fruit With Visible/Near Infra-Red Spectrum

Continuation In Part Application-Continuation In Part Application

This is a Continuation In Part Application copending from the nonprovisional
parent application 09/524,329 entitled AN APPARATUS AND METHOD FOR
MEASURING AND CORRELATING CHARACTERISTICS OF FRUIT WITH
VISIBLE/NEAR INFRA-RED SPECTRUM to Ozanich as filed March 13, 2000. The
applicant requests prosecution pursuant to 37 C.F.R. 1.53(b) and 1.78 and 35 U.S.C. 120.
New matter herein is added, for examination convenience, commencing with page 56
which follows the last line of the Detailed Description of the original application and
precedes the claims.

Field of the Invention-Field of the Invention

The present disclosure relates generally to the use of the combined visible and
near infra red spectrum in an apparatus and method for measuring physical parameters,
e.g., firmness, density and internal and external disorders, and chemical parameters, e.g.,
molecules containing O-H, N-H and C-H chemical bonds, in fruit and correlating the
resulting measurements with fruit quality and maturity characteristics, including Brix,
acidity, density, pH, firmness, color and internal and external defects to forecast
consumer preferences including taste preferences and appearance, as well as harvest,
storage and shipping variables. With the present apparatus and method, the interior of a

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1 sample, e.g., fruit including apples, is illuminated and the spectrum of absorbed and
2 scattered light from the sample is detected and measured. Prediction, calibration and
3 classification algorithms are determined for the category of sample permitting correlation
4 between the spectrum of absorbed and scattered light and sample characteristics, e.g.,
5 ~~fruit~~ fruit quality and

10 **2. At page 2 of the CIP Replacement Application:**

11 **Background of the Invention** Background of the Invention

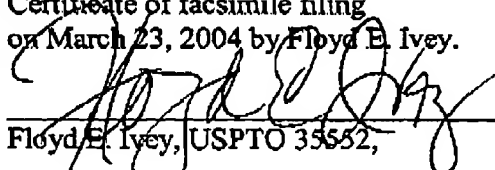
12 The embodiments disclosed herein has a focus on combined visible and near-infrared
13 (NIR) spectroscopy and its modes of use, major issues in the application of NIR to the
14 measurement of O-H, N-H and C-H containing molecules that are indicators of sample
15 quality including fruit quality and in particular tree fruit quality.

16 ~~Near-Infrared Spectroscopy Background:~~ Near-Infrared Spectroscopy Background:

17 Near-infrared spectroscopy has been used since the 1970's for the compositional analysis
18 of low moisture food products. However, only in the last 10-15 years has NIR been
19 successfully applied to the analysis of high moisture products such as fruit. NIR is a form
20 of vibrational spectroscopy that is particularly sensitive to the presence of molecules
21 containing C-H (carbon-hydrogen), O-H (oxygen-hydrogen), and N-H (nitrogen-
22 hydrogen) groups. Therefore, constituents such as sugars and starch (C-H), moisture,
23 alcohols and acids (O-H), and protein (N-H) can be quantified in liquids, solids and
24 slurries. In addition, the analysis of gases (e.g., water vapor, ammonia) is possible. NIR
25 is not a trace analysis technique and it is generally used for measuring components that

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1 are present at concentrations greater than 0.1%.

2 ~~Short-Wavelength NIR vs. Long-Wavelength NIR: Short-Wavelength NIR vs. Long-~~
3 ~~Wavelength NIR:~~ NIR has traditionally been carried out in the 1100-2500 nm region of
4 the electromagnetic spectrum. However, the wavelength region of ~700-1100 nm (short
5 wavelength-NIR or SW-NIR) has been gaining increased attention. The SW-NIR region
6 offers numerous advantages for on-line and *in-situ* bulk constituent analysis. This portion
7 of the NIR is accessible to low-cost, high performance silicon detectors and fiber optics.
8 In addition, high intensity laser diodes and low-cost light emitting diodes are becoming
9 increasingly available at a variety of NIR wavelength outputs.

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12 **3. At page 7 of the CIP Replacement Application:**

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14 transformation and some type of spectral normalization. All of these methods for relating
15 NIR spectra to chemical or physical properties are well known to those practiced in the
16 art of NIR spectroscopy.

17 The foregoing patents and printed publications are provided herewith in an
18 Information Disclosure Statement in accordance with 37 CFR 1.97.

19 **Summary of the Invention** Summary of the Invention

20 Research groups around the world continue to explore the applications of near infrared
21 spectroscopy to tree fruit. The apparatus and process disclosed herein is of

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24 **4. At page 11 of the CIP Replacement Application:**

25 Disclosed herein is an apparatus and method for measurement, with the

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1 visible/near-infrared (VIS/NIR) spectroscopic technique for sugar content (also known as
2 Brix or soluble solids, which is inversely related to dry matter content), firmness, acidity,
3 density, pH, color and internal and external defects and disorders. The apparatus and
4 method is successful in measuring one or more such characteristic in apples, grapes,
5 oranges, potatoes and cherries. Demonstrated in this disclosure is the ability to combine
6 chemical and physical property data permitting the prediction of consumer properties,
7 such as taste, appearance and color; harvest variables, such as time for harvest; and
8 storage variables such as prediction of firmness retention and time until spoilage.

9 Brief Description of the Drawings Brief Description of the Drawings

10 The foregoing and other features and advantages of the present disclosure will
11 become more readily appreciated as the same become better understood by reference to
12 the following detailed description of the preferred embodiment and additional
13 embodiments of the disclosure when taken in conjunction with the accompanying
14 drawings, wherein:

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16 5. At page 19 of the CIP Replacement Application:

17 illuminate the sample(s) 30 with detectors 80 receiving input with light detector output 82
18 conveyed as input to spectrometers 170 detectors 200. In this illustration a lens 130 is depicted
19 between the sample 30 and the detector 80. Illustrations 15 and 15A depict in detail, with filter
20 130 and mounting means, a single detector 80.

21 A CPU 172, controlled by computer program, is not depicted in Fig. 10, 10A, 11, 12, 13, 14,
22 14A, 15 or 15A as a person of ordinary skill will appreciate such structure from viewing other
23 drawings presented herein.

24 Detailed Description Detailed Description

25 The apparatus and method disclosed herein is illustrated in Fig. 1 through 8. Fig.

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1 1C, 1D, 1E and 1F are flow diagrams demonstrating the method of this invention. The
2 flow diagram Fig. 1C is representative of all embodiments of this disclosure. The flow
3 diagram Fig. 1D illustrates one or more light sources 120 and multiple channels from
4 light detector 50 through final prediction of sample characteristic. Fig. 1D demonstrates
5 the method and apparatus of this disclosure illustrating the light source(s) 120, which may
6 be lamps 123 or other light sources, which illuminate a sample 30 interior 36, light
7 collection channels 1...n, composed for example of fiber optic fibers 80 or photodetectors
8 255, e.g., light detector 1...n, of the spectra from a sample 30 delivered as input 82 to a
9 spectra measuring device, shown here as spectrometer(s) 1...n. 170. In the preferred
10 embodiment a light source 120 with lamp 123 is external to the spectrometer and is
11 controlled by a CPU 172 which triggers power 125 to the light source 120 lamp 123.
12 Spectrometer 1...n 170 channels output 1...n are converted from analog to digital by A/D
13 converters 1...n 171 and become, for each channel, input to a CPU 172. The CPU 172 is
14 computer program controlled with each step, following the CPU 172 in this flow diagram
15 is representative of a computer program controlled activity. A CPU 172 output is
16 provided for each channel 1...n where the steps of 1) calculation of absorbance spectra
17 173 occurs for each channel 1...n, 2) combine absorbance spectra 174 into a single
18 spectrum encompassing the entire wavelength range detected from the sample by
19 spectrometers 1...n 170, 3) mathematical preprocessing or preprocess 175, e.g.,
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22 **6. At page 54 of the CIP Replacement Application:**

23 120 and the sample, provided, for example by a linear polarization filter or an equivalent
24 as understood by one of ordinary skill in the art; a matching polarization filter is
25 positioned between the at least one illumination detector and the sample, which may be
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provided, for example by a linear polarization filter rotated 90 degrees in relation to the polarization filter between the light source 120 and the sample.

The method described above, which uses wavelengths of both visible radiation (250-699 nm) specifically chosen to include the absorption band for yellow color pigments (250-499nm), red color pigments (500-600 nm) and green pigments or chlorophyll (601-699 nm), as well as NIR (700-1150 nm) radiation to correlate with Brix, firmness, pH, acidity, density, color and internal and external defects can be carried out using a variety of apparatuses.

ADDITIONAL DETAILED DESCRIPTION

Overview of calibration of visible/NIR sensors: Overview of calibration of

visible/NIR sensors:

Required calibration was addressed in the Parent Application 09/524,329, in paragraphs, identified by page/line by pn/ln, as follows: 1/18; 3/17, 22, 28; 4/2; 8/8; 9/4; 9/14; 12/16; 16/8; 22/5; 31/21; 33/19; 39/10; 43/4; 47/1; 52/13 etc. Calibration of spectroscopic maturity and quality sensors involves building algorithms that relate the visible and near infrared spectrum of an individual fruit or vegetable to one or more of the following: Brix (including, but not limited to sugar content, or sweetness, or soluble solids content); acidity (including but not limited to total acidity, or sourness, or malic acid content or citric acid content or tartaric acid content); pH;

7. At page 55 of the CIP Replacement Application:

Integrating visible/NIR sensors with packing, sorting and conveyance systems and synchronizing data acquisition with product location/position to optimize collection

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1 of sample data, and reference and standardization data.

2 Integrating visible/NIR sensors with packing, sorting and conveyance systems and
3 synchronizing data acquisition with product location/position to optimize collection of
4 sample data, and reference and standardization data.

5 Sensing sample data including the presence or absence of a sample was addressed in the
6 parent in paragraphs, identified by page/line by pn/ln, as follows: 20/20; 36/8 etc. Using
7 spectroscopic sensors for measuring fruits and vegetables while in motion on a sample
8 conveyor 295 system in sorting and packing warehouses is illustrated in Fig. 10 and Fig.
9 10A and is done as follows: The presence or absence of a sample 30 and the
10 position/location of the sample 30 relative to the point of spectrum measurement is
11 determined using one or more of the following means: 1) sample 30 position
12 determination means and or sample conveyor 295 position determination means,
13 provided for example by an encoder or pulse generator 330, as

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18 **8. At page 56 of the CIP Replacement Application:**

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20 Obtaining a spectrum of the lamp(s) for determining reference light output and
21 obtaining baseline dark current spectra from detector(s). Both reference and dark
22 spectra are used with sample spectrum to calculate the product's absorbance
23 spectrum.

24 Obtaining a spectrum of the lamp(s) for determining reference light output and obtaining
25 baseline dark current spectra from detector(s). Both reference and dark spectra are used

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1 with sample spectrum to calculate the product's absorbance spectrum.

2 Reference to reference, baseline and dark spectra was addressed in the parent in
3 paragraphs, identified by page/line by pn/ln, as follows: 12/18; 39/10; 52/14 etc. The
4 reference measurements to account for changes in light source intensity or color output
5 can be obtained using a reference light transmission means 320, e.g., a fiber-optic bundle
6 which may be furcated, a light pipe or other means of transmitting light, with a common
7 end 322 providing input to a reference spectrometer 170, and, where

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11 **9. At pages 60-63 of the CIP Replacement Application:**

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13 A CPU 172, controlled by computer program, is not depicted in Fig. 10, 10A, 11, 12 or
14 13 as a person of ordinary skill will appreciate such structure from viewing other
15 drawings presented herein.

16 ~~**Achieving whole product measurement (minimizing errors due to localized**~~
17 ~~**measurement):**~~

18 **Achieving whole product measurement (minimizing errors due to localized**
19 **measurement).**

20 To improve the measurement of the entire product, two or more light sources 120 lamps
21 123 and/or detection 80 points are used. The product can be measured rolling or not
22 rolling with a rolling measurement generally improving whole product measurement,
23 while a non-rolling measurement provides better accuracy and introduces less spectral
24 noise due to movement.

25 As a single fruit or vegetable sample 30 passes by the point of spectrum acquisition,

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1 multiple spectra are acquired, each spectrum representing a different measurement
2 location or area on the product.

3 ~~Optimizing signal-to-noise and accuracy with small and large size product:~~

4 Optimizing signal-to-noise and accuracy with small and large size product. One or more
5 means may be used to determine the size or weight of the individual fruit or vegetable
6 sample 30. Means for determining product size includes, but is not limited to 1) a
7 separately determined weight or mass using sensors common to the industry, 2) utilizing
8 the color sorter or defect sorter data (e.g., from camera or CCD images), 3) utilizing other
9 size sensors based on magnetic, inductive, light reflectance or multiple light beam
10 curtains, common to other industries. The relative size of the sample 30 can then be used
11 to adjust the hardware spectrum acquisition parameters or the amount of light (by varying
12 the aperture 310 size) to provide an improved signal-to-noise ratio spectrum for large
13 samples 30 and/or to prevent detector 80 saturation by light for small product sample 30,
14 e.g., detector 80 exposure or integration time can be set for longer time periods for large
15 product samples 30 and for shorter time periods for small product.

16 ~~Improving accuracy by inspection of multiple individual spectra collected from a~~
17 ~~single product and removing poor quality or "outlier" spectra. Then, calculating~~
18 ~~the absorbance spectrum from the raw data collected for dark, reference and~~
19 ~~sample:~~

20 Improving accuracy by inspection of multiple individual spectra collected from a single
21 product and removing poor quality or "outlier" spectra. Then, calculating the absorbance
22 spectrum from the raw data collected for dark, reference and sample.

23 Each individual spectrum from the series of spectra acquired for each individual product
24 sample 30 are then inspected by a computer program or programmed hardware. Poor
25 quality spectra are deleted from this batch of spectra and the remaining spectra are used

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1 for constituent or property prediction. The retained spectra of the product are combined
2 with the appropriate reference and dark current measurements to produce an absorbance
3 spectrum as follows:

4 Absorbance Spectrum = $-\log_{10} [(sample\ intensity\ spectrum - sample\ dark\ current$
5 $spectrum) / (reference\ intensity\ spectrum - reference\ dark\ current\ spectrum)]$ i.e. the
6 absorbance spectrum is equal to the negative logarithm (base 10) of the ratio of the dark
7 current corrected sample spectrum to the dark current corrected reference spectrum.

8 All of the absorbance spectra for each product sample 30 can then be combined to
9 produce a mean or average absorbance spectrum of the product sample. This average
10 absorbance spectra can then be used to compute the component or property of interest
11 based on a previously stored calibration algorithm. Alternatively, each absorbance
12 spectrum can be used individually with a previously stored calibration algorithm to
13 compute multiple results of the component or property of interest for an individual
14 product, followed by determination of the average or mean component or property value
15 computed by summing all of the values and dividing the resultant sum by the number of
16 absorbance spectra used.

17 Method for measuring samples and importance of linking location on product
18 where visible/NIR data was collected with the same location that will be measured
19 by the laboratory reference technique.

20 Method for measuring samples and importance of linking location on product where
21 visible/NIR data was collected with the same location that will be measured by the
22 laboratory reference technique.

23 Calibration is performed as follows: 1) Spectra of product sample 30 are measured and
24 absorbance spectra (corrected for reference and dark current) are stored, 2) Standard
25 laboratory measurements (which are often destructive) are made on the product sample

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1 30. Note: it is important to the success of the NIR method that the portion of the sample
2 30 that is interrogated between the light source(s) 120 lamps 123 and light collection(s)
3 detectors, e.g., light detectors 80, leading to the spectrometer(s) 170 detectors 200 is the
4 same as that portion measured by the standard laboratory technique.

5 For many sample conveyors 295 that are used for whole fruit and vegetable sorting and
6 packing operations, the product can be transported past the NIR measurement location
7 rolling or not rolling. If absorbance spectra are collected from the product as it is rolling,
8 the exact location of any one measurement (one spectrum) is not usually known, and
9 therefore the entire product (as opposed to one localized spot) must be analyzed for the
10 component or property of interest. If calibration algorithms are constructed in this way
11 (using measurements of rolling product), all of the retained spectra for that individual
12 product are averaged to produce an average absorbance spectrum and the total product
13 component or property is assigned to this one absorbance spectrum.

14 Because most fruits and vegetable are heterogeneous and vary in component level with
15 location, it is preferable to develop a calibration model on product sample 30 that is not
16 rolling so that each acquired spectrum is from a known physical location on the product
17 sample 30. Then, laboratory measurements are made on the same portion of product
18 sample 30 that spectra were taken from. When this procedure is used, a whole fruit or
19 vegetable sample 30 may be separated, e.g., cut or sliced, into smaller sub-portions prior
20 to laboratory analysis. These smaller sub-portions each correspond to NIR data collected
21 over the same locations within the product sample 30; the time period of NIR data
22 acquisition can be adjusted to shorter or longer times, corresponding to the measurement
23 of smaller or larger product samples 30, respectively. In this case, each sub-portion of the
24 product sample 30 will have one or more spectra associated with that particular location.
25 The laboratory determined component or property is then assigned to each spectrum or

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1 spectra from that particular location.

2 ~~Mathematical processing is performed on absorbance spectra prior to conducting~~
3 ~~statistical correlation analysis and calibration model building.~~

4 Mathematical processing is performed on absorbance spectra prior to conducting
5 statistical correlation analysis and calibration model building.

6 Absorbance spectra are pre-processed using a bin and smooth function. Partial least
7 squares analysis (or variants thereof such as piecewise direct standardization) are then
8 used to relate the processed absorbance spectrum to the assigned component and property
9 values such as Brix, acidity, pH, firmness, color, internal or external disorder severity and
10 type, and eating quality.

11 ~~Method to minimize the number of samples needed to develop a calibration model.~~

12 Method to minimize the number of samples needed to develop a calibration model.

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